and delineate active neurons permitting automatic analysis of activity in neurons via in vivo calcium imaging.

3254-Plat
Overcoming Reverse Rate Dependence in Ventricular Cell Models
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Reverse rate dependence (RRD) is a problematic property of antiarrhythmic drugs that prolong the action potential duration (APD). The prolongation caused by RRD agents will increase at slow rates, resulting in both reduced arrhythmia suppression at fast rates and increased arrhythmogenesis at slow. The opposite property, forward rate dependence (FRD), would theoretically overcome these parallel problems, yet FRD antiarrhythmics remain elusive. Moreover, there is evidence that RRD is an intrinsic property of perturbations to the action potential (AP). We have addressed the possibility of FRD by performing a comprehensive analysis of 13 ventricular myocyte models. By simulating populations of myocytes with varying properties and analyzing population results statistically, we were able to simultaneously predict the rate-dependent effects on the APD of changes in any of an average of 40 parameters per model. The analysis produced several important results. First, while models often display RRD, a variety of ion current perturbations do in fact produce FRD. Second, additional simulations of FRD perturbations provide mechanistic insight into how FRD behavior can be produced. For instance, increasing L-type calcium conductance (GCaL) is FRD when accompanied by concomitant, indirect, rate-dependent changes in slow delayed rectifier current (IKs). Third, comparisons of results between models revealed that changes in IKs are almost always RRD whereas changes in GCaL or the Na-K pump can potentially be FRD. Fourth, the general capacity for FRD correlates strongly with the degree of rate-dependent change in AP shape. Models that display minimal changes in AP shape with rate have little capacity for FRD whereas models with large shape changes have considerable FRD potential. Overall, this study provides new insight into the determinants of APD rate dependence and illustrates a strategy for the design of potentially beneficial antiarrhythmic drugs.

3255-Plat
Elucidating Metabolic Variability in Isogenic Microbial Populations Arising due to Noise in Protein Expression
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Members of a population of isogenic microbes vary in their protein expression states because of stochastic gene expression. Advances in single-cell proteomics on model organisms like E. coli and S. cerevisiae are uncovering this heterogeneity in protein copy numbers. Here we use genome-scale flux balance models to study the effect of heterogeneity in protein expression on metabolic behavior. We predict wide distribution in specific growth rates among the members of the population in accordance with recent single-cell growth rate measurements. Using flux balance analysis along with principal component analysis enables us to identify sub-populations which differ in their metabolic pathway usage. In case of E. coli grown in a minimal medium, we predict presence of slow-growing acetate secreting cells, fast-growing CO2 secreting cells and shifting preference between glycolysis and ED pathway to metabolize glucose. Preliminary population-level measurements support our prediction of acetate secreting cells in aerobic growth conditions. We also find that variability in expression of few genes may be sufficient to capture most of the metabolic variability of the entire population. We extend this study to S. cerevisiae for which the requisite data is becoming available.

3256-Plat
Computational Modeling of Granuloma Formation in Tuberculosis Yields Insights into both Infection and Treatment
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Tuberculosis (TB), a disease caused by Mycobacterium tuberculosis (Mtb), results in 1-2 million deaths/year. Disease control is hampered by our limited understanding of the relevant biology and by development of antibiotic resistance. Granulomas, organized collections of immune cells and bacteria that form in lungs, are central features of TB and serve as sites of host-pathogen interaction. Cytokines influence the behavior of immune cells, directing granuloma function and maintenance. Granuloma structure helps determine bacterial phenotypes, antibiotic distribution and efficacy. There is a critical need for an in silico platform to provide cost-effective means of understanding the immune response during Mtb infection and testing and optimizing new treatment strategies.
We developed a multi-scale computational model of the immune response to Mtb. Molecular, cellular and tissue behavior over minutes to years can be computed, validated with in vitro and in vivo data, and used to understand and predict system behavior. The model incorporates tuneable resolution, allowing us to vary the aspect and level of detail for virtual experiments. We present two examples of model use. First, we study how concentrations of a pro-inflammatory cytokine, tumor necrosis factor-α, and an anti-inflammatory cytokine, interleukin-10, control granuloma formation and function. We find that a balance of concentrations defines a granuloma environment that may benefit both host and pathogen. Second, we explore the role of granuloma structure in antibiotic distribution and action. Antibiotic concentration gradients form within granulomas and could contribute to development of resistance. We compare dosing regimens of two first-line antibiotics, isoniazid and rifampicin, and demonstrate that intermittent high doses are less effective than daily low dose regimens. Our computational approach represents a critical step towards understanding the complex phenomena involved in Mtb infection and developing successful treatment strategies.

Platform: Cardiac Muscle II

3257-Plat
E-C Coupling Alterations and Spontaneous Activity in Mice Carrying Cardiac Troponin T Mutations
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Ca^{2+} handling abnormalities are an early-onset pathogenic element in HCM. Here we characterize pro-arrhythmogenic changes in E-C coupling that occur in intact trabeculae and cardiomyocytes from cTnT mutant mouse models of HCM (R92Q, E163R and Δ160E) and test the effects of specific pharmacological interventions. Compared to WT, R92Q trabeculae ($[Ca^{2+}]_o$ 2 mM, 30°C) showed (i) preserved peak isometric twitch tension and prolonged relaxation kinetics associated to decreased SERCA levels, (ii) faster mechanical restitution, further accelerated by isoproterenol (Iso) 100nM, (iii) decreased Ca^{2+}-recirculation fraction markedly increased by Iso (iv) frequent after-contractions or regular spontaneous beats during stimulation pauses that increased in response to Iso. Compared to WT, R92Q cardiomyocytesshowed (i) prolonged action potentials associated with ionic current remodeling, (ii) slower rate of Ca^{2+}transient decay, (iii) elevated diastolic $[Ca^{2+}]_i$, (iv) spontaneous Ca^{2+} waves during stimulation pauses. In R92Q preparations, the late-Na^+ current blocker Ranolazine (Ran 10 μM) (i) reduced the rate of spontaneous beats and spontaneous Ca^{2+} waves, (ii) hastened Ca^{2+} transient kinetics and reduced diastolic Ca^{2+}, (iii) reduced and reversed the acceleration of mechanical restitution and the increase in Ca^{2+} recirculation fraction induced by Iso.Compared to R92Q, occurrence of spontaneous contractions was similar in E163R but less pronounced in Δ160E. Iso and Ran showed similar effects in all three mouse models, in spite of some quantitative differences. The results are consistent with those recently reported in human HCM myocytes (Coppini et al, Circulation 2013) and suggest that remodeling and dysfunction of NCX and RyR2 contribute to the pro-arrhythmogenic E-C coupling abnormalities observed in HCM.

3258-Plat
Familial Hypertrophic Cardiomyopathy: Unequal Expression of Mutant and Wildtype Myosin in Individual Myocytes as Trigger for Functional Impairment of the Heart?
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In Familial Hypertrophic Cardiomyopathy (FHC) the direct effects of disease-causing mutations in sarcomeric proteins on sarcomere function are still largely unknown. The current hypothesis is that FHC-myofilament mutations cause an increase in calcium-sensitivity, force and ATPase.